Design, Synthesis and Biological Evaluation of Some Novel 3-Methylyquinoxaline-2-Hydrazone Derivatives

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Abstract
Alzheimer’s disease is a chronic and progressive neurodegenerative disease which occurs due to lower levels of acetylcholine neurotransmitters, and results in a gradual decline in memory and other cognitive processes. Acetylcholinesterase and butyrylcholinesterase have been reported to be the primary regulators of the acetylcholine levels in the brain. Evidence shows that acetylcholinesterase activity decreases in Alzheimer’s disease, while activity of butyrylcholinesterase elevate in advanced Alzheimer’s disease, which suggests a key involvement of butyrylcholinesterase in acetylcholine hydrolysis during Alzheimer’s disease symptoms. In order to sustain the level of remaining acetylcholine, acetylcholinesterase and butyrylcholinesterase inhibitors may be used. Therefore, inhibiting the activity of butyrylcholinesterase may be an effective way to control Alzheimer’s disease associated disorders. In this study, eleven 3-methylyquinoxaline-2-hydrazones were synthesized from the reactions of 3-methylquinoxaline-2-hydrazide with different substituted aromatic ketones and aromatic aldehyde. All the newly synthesized compounds have been characterized on the basis of IR, 1H-NMR and 13C-NMR spectral data as well as physical data. All the synthesized compounds were biologically evaluated against cholinesterases (acetylcholinesterase and butyrylcholinesterase). Compounds 2-12 were found to be a good selective inhibitor for acetylcholinesterase and butyrylcholinesterase. Among the series, compounds 6 (IC50 =170 ± 30 µg/mL) and 10 (IC50 =180 ± 10 µg/mL) were found to be the most active inhibitors against acetylcholinesterase, while compounds 2 (IC50 =780 ± 10 µg/mL), 5 (IC50 =550 ± 10 µg/mL) and 6 (IC50 =790 ± 10 µg/mL) were found to be most active inhibitor against butyrylcholinesterase. The IC50 values for all the synthesized compounds were lower than standard, eserine (IC50 =70 ± 20 µg/mL). Their considerable activity of butyrylcholinesterase and butyrylcholinesterase inhibitory activities makes them a good candidate for the development of selective acetylcholinesterase and butyrylcholinesterase inhibitors.

Keywords: Alzheimer's disease; Acetylcholine; 3Methylquinoxalin; Acetylcholinesterase; Butyrylcholinesterase; Eserine; Hydrazones

Introduction
Heterocyclic compounds represent an important class of biological active molecules [1]. Specifically those containing quinoxaline derivatives have evoked considerable attention in recent years. Quinoxaline, or 1,4-benzo[pyrazine is an important structural unit among nitrogen-containing heterocyclic compounds. Quinoxalines are, in general, easy to prepare and numerous derivatives have been reported in the literature because of their biological activity, specifically as antimicrobial [2-8], antibacterial [9-11], anti-cancer [12], antiaminoceptive [13], anti-inflammatory [14,15] anti-viral [16-18], antimalaria [19] agents. They possess well known biological activities including AMPA/GlyN receptor antagonist [20], antihistaminic agents [21], anti-trypanosomal activity [22], anti-herps, trypanocida, antimalarial activity [23], Ca++ uptake/release inhibitor [24], and inhibitor of vascular smooth muscle cell proliferation. Quinoxaline derivatives constitute the basis of many insecticides, fungicides, herbicides, as well as being important in human health and as receptor antagonists. Although rarely described in nature, synthetic quinoxaline moiety is a part of number of antibiotics such as echinomycin [25], levomycin and actinomycin which are known to inhibit the growth of Gram-positive bacteria and also active against various transplantable tumors [5,26]. In addition, quinoxaline derivatives are reported for their application in dyes, efficient electroluminescent materials, organic semiconductors, and DNA cleaving agents [27].

Alzheimer’s disease is a common form of dementia in which severe loss of cholinergic cell occurs, which subsequently leads to low levels of the neurotransmitter acetylcholine in brain. Acetylcholinesterase is the key enzyme involved in the metabolic hydrolysis of acetylcholine. This observation led to the introduction of the acetylcholinesterase inhibitors to prolong the duration of action of acetylcholine [28]. Another enzyme, butyrylcholinesterase, expressed in selected areas of the central and peripheral nervous systems, is also capable of hydrolyzing acetylcholine [29]. The management of Alzheimer’s disease has been a long-standing challenge and area of interest [30]. Despite the long history of the disease, there are currently very few drugs used clinically for the management of Alzheimer’s disease [31]. This work was therefore designed to synthesize new quinoxaline compounds carrying the hydrazone functional group at the 2-position, elucidate their structures, in addition the compounds were evaluated against cholinesterases (acetylcholinesterase and butyrylcholinesterase).

Materials and Methods
Melted points were determined with open capillary tube on a Gallenkamp (variable heater) melting point apparatus and were uncorrected. Infrared spectra were recorded as KBr pellets on a Buck Spectrometer. The 1H-NMR and 13C-NMR was run on a Bruker 600 MHz spectrometer (δ in ppm relative to Me4Si) at the Department of Chemistry, Portland state University, Portland USA. The purity of the synthesized compounds was determined by thin layer chromatography (TLC) on silica gel TLC plates.

Abstract
Alzheimer’s disease is a chronic and progressive neurodegenerative disease which occurs due to lower levels of acetylcholine neurotransmitters, and results in a gradual decline in memory and other cognitive processes. Acetylcholinesterase and butyrylcholinesterase have been reported to be the primary regulators of the acetylcholine levels in the brain. Evidence shows that acetylcholinesterase activity decreases in Alzheimer’s disease, while activity of butyrylcholinesterase elevate in advanced Alzheimer’s disease, which suggests a key involvement of butyrylcholinesterase in acetylcholine hydrolysis during Alzheimer’s disease symptoms. In order to sustain the level of remaining acetylcholine, acetylcholinesterase and butyrylcholinesterase inhibitors may be used. Therefore, inhibiting the activity of butyrylcholinesterase may be an effective way to control Alzheimer’s disease associated disorders. In this study, eleven 3-methylyquinoxaline-2-hydrazones were synthesized from the reactions of 3-methylquinoxaline-2-hydrazide with different substituted aromatic ketones and aromatic aldehyde. All the newly synthesized compounds have been characterized on the basis of IR, 1H-NMR and 13C-NMR spectral data as well as physical data. All the synthesized compounds were biologically evaluated against cholinesterases (acetylcholinesterase and butyrylcholinesterase). Compounds 2-12 were found to be a good selective inhibitor for acetylcholinesterase and butyrylcholinesterase. Among the series, compounds 6 (IC50 =170 ± 30 µg/mL) and 10 (IC50 =180 ± 10 µg/mL) were found to be the most active inhibitors against acetylcholinesterase, while compounds 2 (IC50 =780 ± 10 µg/mL), 5 (IC50 =550 ± 10 µg/mL) and 6 (IC50 =790 ± 10 µg/mL) were found to be most active inhibitor against butyrylcholinesterase. The IC50 values for all the synthesized compounds were lower than standard, eserine (IC50 =70 ± 20 µg/mL). Their considerable activity of butyrylcholinesterase and butyrylcholinesterase inhibitory activities makes them a good candidate for the development of selective acetylcholinesterase and butyrylcholinesterase inhibitors.

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compounds was routinely checked by TLC on silica gel G plates using n-hexane/ethyl acetate (1:1, v/v) solvent system and the developed plates were visualized by UV light. All reagents used were obtained from Sigma-Aldrich Chemical Ltd.

**Synthesis of 3-methylquinoxaline-2-(1H)-one**

O-phenylenediamine (20 g 0.10 M) and ethyl pyruvate (22 g 0.10 M) in 200 ml of absolute ethanol was heated for 30 minutes on oil bath. The reaction mixture was allowed to cool to give some silvery white crystals which were collected by filtration, washed and purified by recrystallization from ethanol.

**Synthesis of 2-hydrazinyl-3-methyl-1,2-dihydroquinoline 1**

3-methyl quinoxaline-2-(1H)-one (10 g. 0.0625 mol) was added to a mixture hydrazine hydrate and 20 ml of water. The resulting mixture was refluxed for 4 hours. The reaction mixture was allowed to cool to room temperature to afford a brownish-yellow solid precipitate which was filtered, dried and recrystallized from ethanol.

**General procedure for the reaction of 2-hydrazinyl-3-methyl-1,2-dihydroquinoline with substituted aromatic ketones**

2-hydrazinyl-3-methyl-1,2-dihydroquinoline (1.0 g. 5.67 mmol) and various substituted aromatic ketones (5.67 mmol) were added to glacial acetic acid (10 ml) in a round bottom flask and refluxed at 120°C for 3 hours. The reaction mixture was cooled and poured into crushed ice with continuous stirring to obtain a solid product which was filtered and dried. Recrystallization from DMF/water afforded the hydrazones 2-11.

**Spectral data synthesized compounds**

3-methylquinoxaline-2(1H)-one: % yield: 76.44%; Melting point: 245-247°C lit. 246°C [15-16]; IR KBr (cm\(^{-1}\)): 3103 (C-H sp\(^3\) str.), 1602 (C=C aromatic str.), 1660 (C=N str.), 2866 (C-H sp\(^3\) str.); 1568 (N-H bend), 1665 (C=N str.); 2966 (C-H sp\(^3\) str.), 3448 (N-H str.), 3308 (C=N str.); 1608 (C=C aromatic str.), 1598 (N-H bend).

**2-(E)-2-(1-((3-butanoyl)ethylideneco)hydrazinyl)-3-methylquinoxaline (3):** % yield: 78.40%; Melting point: 252-254°C; IR KBr (cm\(^{-1}\)): 3442 (N-H str.), 2910 (C-H sp\(^3\) str.), 1605 (C=C aromatic str.), 1664 (C=N str.), 1598 (N-H bend).

**N-H NM- (DMSO-d\(_6\)):** 10.43 (broad s, 1H, hydrazine NH), 7.95 (d, 2H, aromatics protons), 7.93 (d, 1H, aromatic proton); 7.90 (d, 1H, aromatic proton); 7.76 (t, 1H, aromatic protons); 7.55 (t, 1H, aromatics proton); 7.53 (t, 2H, aromatics protons); 2.94 (s, 3H, methyl proton). 2.40 (s, 3H, methyl proton).

**13C-NMR (DMSO-d\(_6\)):** 163 ppm, 145 ppm, 135 ppm, 127 ppm, 125 ppm, 124 ppm, 17 ppm.

(E)-2-methyl-3-(2-(1-phenylthiophenylidenedehydrazinyl)quinoxaline (2): % yield: 75.40%; Melting point: 252-254°C; IR KBr (cm\(^{-1}\)): 3435 (N-H str.), 2899 (C-H sp\(^3\) str.), 1602 (C=C aromatic str.), 1665 (C=N str.), 1564 (N-H bend), 1008 (N-N str.).

**1-H NM- (DMSO-d\(_6\)):** 10.43 (broad s, 1H, hydrazine NH), 7.95 (d, 2H, aromatics protons), 7.93 (d, 1H, aromatic proton); 7.90 (d, 1H, aromatic proton); 7.76 (t, 1H, aromatic protons); 7.55 (t, 1H, aromatics proton); 7.53 (t, 2H, aromatics protons); 2.94 (s, 3H, methyl proton). 2.40 (s, 3H, methyl proton).

**13C-NMR (DMSO-d\(_6\)):** 168 ppm, 163 ppm, 145 ppm, 137 ppm, 135 ppm, 131 ppm, 127 ppm, 125 ppm, 124 ppm, 17 ppm.

(E)-2-(2-(1-(4-bromophenyl)ethylidene)hydrazinyl)-3-methylquinoxaline (3): % yield: 78.40%; Melting point: 252-254°C; IR KBr (cm\(^{-1}\)): 3442 (N-H str.), 2910 (C-H sp\(^3\) str.), 1605 (C=C aromatic str.), 1664 (C=N str.), 1598 (N-H bend).

**N-H NM- (DMSO-d\(_6\)):** 10.43 (broad s, 1H, hydrazine NH), 7.60-7.90 (m, 4H, aromatics protons); 7.67 (d, 1H, aromatic protons); 7.43 (J=8, 8, 2) (t, 1H, aromatic protons); 7.27 (J=8, 8, 2) (t, 1H, aromatic protons); 7.36 (d, 1H, aromatics protons) 2.94 (s, 3H, methyl proton), 2.40(s, 3H, methyl proton).

**13C-NMR (DMSO-d\(_6\)):** 168 ppm, 164 ppm, 145 ppm, 135 ppm, 133 ppm, 130 ppm, 127 ppm, 125 ppm 124 ppm, 123 ppm, 17 ppm.

(E)-2-(2-(1-(2-fluorophenyl)ethylidene)hydrazinyl)-3-methylquinoxaline (4): % yield: 71.10%; Melting point: 207-209°C; IR KBr (cm\(^{-1}\)): 3435 (N-H str.), 2966 (C-H sp\(^3\) str.), 1608 (C=C aromatic str.), 1662 (C=N str.), 1564 (N-H bend), 1008 (N-N str.).

**N-H NM- (DMSO-d\(_6\)):** 10.46 (broad s, 1H, hydrazine NH), 7.60-7.94 (m, 4H, aromatic protons); 7.67 (d, 1H, aromatic protons); 7.48 (J=8, 8, 2) (t, 1H, aromatic protons); 7.27 (J=8, 8, 2) (t, 1H, aromatic protons); 7.36 (d, 1H, aromatics protons) 2.95 (s, 3H, methyl proton). 2.43 (s, 3H, methyl proton).

**13C-NMR (DMSO-d\(_6\)):** 168 ppm, 159 ppm, 145 ppm, 135 ppm, 133 ppm, 130 ppm, 127 ppm, 124 ppm, 118 ppm, 17 ppm.

(E)-2-(2-(1-(4-fluorophenyl)ethylidene)hydrazinyl)-3-methylquinoxaline (5): % yield: 85.40%; Melting point: 250-252°C; IR KBr (cm\(^{-1}\)): 3458 (N-H str.), 2850 (C-H sp\(^3\) str.), 1618 (C=C aromatic str.), 1660 (C=N str.), 1548 (N-H bend).

**N-H NM- (DMSO-d\(_6\)):** 10.46 (broad s, 1H, hydrazine NH), 7.27 (m, 2H, aromatic protons); 7.67 (d, 1H, aromatic protons); 7.48 (J=8, 8, 2) (t, 1H, aromatic protons); 7.27 (J=8, 8, 2) (t, 1H, aromatic protons); 7.19 (m, 2H, aromatics protons), 2.93 (s, 3H, methyl proton), 2.41 (s, 3H, methyl proton).

**13C-NMR (DMSO-d\(_6\)):** 168 ppm, 165 ppm (C-F), 163 ppm, 145 ppm, 135 ppm, 134 ppm, 133 ppm, 129 ppm, 127 ppm, 124 ppm, 117 ppm, 17 ppm.

(E)-2-(2-(1-(2,4-dibromophenyl)ethylidene)hydrazinyl)-3-methylquinoxaline (6): % yield: 77.35%; Melting point: 183-184°C; IR KBr (cm\(^{-1}\)): 3103 (C-H sp\(^3\) str.), 2897 (C-H sp\(^3\) str.), 1605 (C=C aromatic str.), 1669 (C=N str.), 1568 (N-H bend).

**N-H NM- (DMSO-d\(_6\)):** 10.46 (broad s, 1H, hydrazine NH), 7.27 (m, 3H, aromatic protons).
2H, aromatic protons); 7.67 (d, 1H, aromatic proton); 7.48 (J=8, 8, 2) (t, 1H, aromatic proton); 7.69 (J=8, 8, 2) (m, 2H, aromatic protons); 7.94 (m, 1H, aromatics protons) 2.94 (s, 3H, methyl proton), 2.40 (s, 3H, methyl proton).

\[ ^{13}C\text{-NMR (DMSO-d}_6\text{):} \]
168 ppm, 164 ppm, 145 ppm, 135 ppm, 134 ppm, 133 ppm, 130 ppm, 127 ppm, 124 ppm, 123 ppm, 17 ppm, 15 ppm.

(E)-4-(1-(2-(3-methylquinoxalin-2-yl)hydrazono)ethyl)aniline (7): % yield: 45.40%; Melting point: 252-254°C; IR KBr (cm\(^{-1}\)): 3436 (N-H str.), 2966 (C-H str.), 1608 (C=C Aromatic str.), 1600 (C=N str.), 1564 (N-H bend), 1008 (N-N str.)

\[ ^{13}H\text{-NMR (DMSO-d}_6\text{):} \]
10.46 (broad s, 1H, hydrazine NH), 7.27 (m, 2H, aromatic protons); 7.67 (d, 1H, aromatic proton); 7.64 (d, 2H, aromatic protons); 6.88 (d, 2H, aromatic protons); 7.94 (m, 1H, aromatics protons), 5.48 (s, 2H, \(\text{NH}_2\)), 2.94 (s, 3H, methyl proton), 2.40 (s, 3H, methyl proton).

\[ ^{13}C\text{-NMR (DMSO-d}_6\text{):} \]
168 ppm, 164 ppm, 145 ppm, 135 ppm, 134 ppm, 133 ppm, 130 ppm, 127 ppm, 124 ppm, 123 ppm, 17 ppm, 15 ppm.

2-(2-(3-methylquinoxalin-2-yl)hydrazono)-1H-indene-1,3(2H)-dione (8): % yield: 52.30%; Melting point: 219-221°C; IR KBr (cm\(^{-1}\)): 3436 (N-H str.), 2966 (C-H str.), 1608 (C=C Aromatic str.), 1600 (C=N str.), 1564 (N-H bend), 1008 (N-N str.)

\[ ^{13}H\text{-NMR (DMSO-d}_6\text{):} \]
10.45 (broad s, 1H, hydrazine NH), 7.93 (d, 1H, aromatic proton); 7.90 (d, 1H, aromatic proton); 7.80 (d, 1H, aromatic proton); 7.76 (t, 1H, aromatic protons); 7.67 (d, 2H, aromatic protons); 7.67 (t, 1H, aromatics proton); 7.61 (t, 1H, aromatic proton), 2.40 (s, 3H, methyl proton)

\[ ^{13}C\text{-NMR (DMSO-d}_6\text{):} \]
168 ppm, 164 ppm, 145 ppm, 135 ppm, 134 ppm, 133 ppm, 130 ppm, 127 ppm, 124 ppm, 123 ppm, 17 ppm, 15 ppm.

2-(1,3-dihydro-2H-inden-2-ylidene)hydrazinyl)-3-methylquinoxaline (11): % yield: 80.20%; Melting point: 249–251°C; IR KBr (cm\(^{-1}\)): 3442 (N-H str.), 2968 (C-H sp\(^3\) str.), 1602 (C=C aromatic str.), 1570(C=N str.) 1008 (N-N str.)

\[ ^{13}H\text{-NMR (DMSO-d}_6\text{):} \]
10.45 (broad s, 1H, hydrazine NH), 7.93 (d, 1H, aromatic proton); 7.90 (d, 1H, aromatic proton); 7.75 (d, 1H, aromatic proton); 7.76 (t, 1H, aromatic protons); 7.45 (d, 2H, aromatics proton); 7.37 (t, 2H, aromatic protons), 3.39 (d, 4H, aliphatic protons); 2.40 (s, 3H, methyl proton)

\[ ^{13}C\text{-NMR (DMSO-d}_6\text{):} \]
163 ppm, 155 ppm, 145 ppm, 141 ppm, 135 ppm, 128 ppm, 127 ppm, 125 ppm, 124 ppm, 38 ppm, 32 ppm, 17 ppm.

1-(2-methylquinoxalin-3-yl)-2-((naphthalen-1-yl)methylene) hydrazine (12): % yield: 85.00%; Melting point: 160-161°C; IR KBr (cm\(^{-1}\)): 3442 (N-H str.), 2900 (C-H sp\(^3\) str.), 1610 (C=C Aromatic str.), 1568(C=N str.)

\[ ^{13}H\text{-NMR (DMSO-d}_6\text{):} \]
10.45 (broad s, 1H, hydrazine NH), 8.55 (s, 1H, hydrazino proton); 8.50 (d, 1H, aromatic proton); 7.98 (t, 1H, aromatic proton); 7.93 (d, 1H, aromatic proton); 7.93 (d, 1H, aromatic proton); 7.90 (d, 1H, aromatic proton); 7.77 (t, 1H, aromatic proton); 7.75 (d, 1H, aromatic proton); 7.76 (t, 1H, aromatic protons); 7.45 (t, 2H, aromatic protons), 2.40 (s, 3H, methyl proton).

\[ ^{13}C\text{-NMR (DMSO-d}_6\text{):} \]
163 ppm, 145 ppm, 143 ppm, 135 ppm, 133 ppm, 130 ppm, 128 ppm, 127 ppm, 125 ppm, 124 ppm, 17 ppm.

**Cholinesterase inhibitory assay**

AChE and BuChE inhibitions were determined spectrophotometrically using acetyl thiocholine iodide (ATChI) and butyrylthiocholine chloride (BuChCl) as substrate, respectively by the modified method of Ellman et al.

In a 96-well plate was added 240 µl of buffer (50 mM Tris-HCl, pH 8.0) and 20 µl of varying concentrations of the test samples (10, 5, 2.5 and 1.25 mg/ml), 20 µl of the enzyme preparation.

\[ \text{Where: } I(%) = \frac{V_o - V_i}{V_o} \times 100 \]

A solution of buffer was used as negative control. All assays were carried out in triplicate. Eserine (+) physostigmine was used as positive control.

The percentage inhibition (I%) of test sample was obtained using the formula:

\[ I(\%) = \frac{V_o - V_i}{V_o} \times 100 \]

Where: I(%)=Percentage inhibition

\[ V_o = \text{enzyme activity in the absence of test sample} \]
\[ V_i = \text{enzyme activity in the presence of test sample} \]

The synthesized compounds 2-12 were subjected to this test.

**Results and Discussion**

**Chemistry**

3-methylquinoxalin-2-one was prepared from the reaction of o-phenylenediamine with ethyl pyruvate in n-butanol (Scheme 1). The
precursor was synthesized from the reaction of 3-methylquinoxaline-2-one with hydrazine hydrate to obtain the 3-methylquinoxaline-2-hydrazine (I). The reaction was carried out by stirring under reflux for four hours. Eleven Nitrogen containing heterocyclic compounds containing 3-methylquinoxaline-2-hydrazone group were synthesized using conventional heating method. These heterocycles include derivatives of Ninhydrin, Curcumin, Isatin, 1-Indanone, Naphthaldehyde and substituted acetophenones (Scheme 2).

The structures of the compounds were partially characterised using Infrared, $^1$H and $^{13}$C Nuclear Magnetic Resonance spectroscopic methods. All the spectroscopic data confirmed the structures of all the compounds synthesized. The diagnostic bands in the IR spectral for the formation of hydrazones bond C=N were observed between 1564 and 1679 cm$^{-1}$. The NH bands appeared between 3409 and 3451 cm$^{-1}$, while the CH-SP$^3$ stretching frequency appeared between 2899 and 2969 cm$^{-1}$. The methyl group appeared in the region of 2.40 and 2.95 ppm in the $^1$H-NMR spectral, the azomethine group CH=N- proton appeared between 8.55 and 10.46 ppm in the $^1$H-NMR spectral.

Biology

Cholinesterase enzymes inhibitory activity: Cholinesterase enzymes inhibitory activity of 2-12 were evaluated using Ellman's assay. Compounds 2-12 were found to be a good selective inhibitor for acetylcholinesterase and butyrylcholinesterase (Table 1). Among the series, compounds 6 ($IC_{50}=170 \pm 30 \mu g/mL$) and 10 ($IC_{50}=180 \pm 10 \mu g/mL$) were found to be the most active inhibitors against acetylcholinesterase, while compounds 2 ($IC_{50}=780 \pm 10 \mu g/mL$), 5 ($IC_{50}=550 \pm 10 \mu g/mL$) and 6 ($IC_{50}=790 \pm 10 \mu g/mL$), were found to be most active inhibitor against butyrylcholinesterase. The $IC_{50}$

\begin{center}
\textbf{Scheme 1:} Reaction of \(o\)-phenylenediamine with ethyl pyruvate in \(n\)-butanol.
\end{center}
values for all the synthesized compounds were lower than standard, eserine (IC_{50}=70 ± 20 µg/mL). Their considerable acetylcholinesterase and butyrylcholinesterase inhibitory activities makes them a good candidates for the development of selective acetylcholinesterase and butyrylcholinesterase inhibitors. All the compounds were found to be highly selective to acetylcholinesterase and butyrylcholinesterase, respectively.

Alzheimer's disease is a chronic and progressive neurodegenerative disease. The biochemical deficits of this disease are caused by reduced levels of acetylcholine due to substantial reduction in the activity of the enzyme choline acetyltransferase, reduced activity of acetylcholinesterase, and by contrast increased activity of butyrylcholinesterase [32]. In order to sustain the level of remaining acetylcholine, acetylcholinesterase and butyrylcholinesterase inhibitors are used. Thus, compounds 2-12 have potential in the treatment of Alzheimer's disease. Even though they were less active than eserine, they may serve as a potential lead compound for the synthesis of more bioactive derivatives.

In addition, butyrylcholinesterase has been implicated to play important role in the development and progressing of Alzheimer's disease. It cleaves the amyloid precursor protein to β-amyloid that will progress to form α-amyloid plaques, which leads to neurodegeneration. Thus, selective butyrylcholinesterase inhibitor have been reported to prevents the formation of α-amyloid plaques [33]. According to Greig et al., selective butyrylcholinesterase inhibitor may be useful in ameliorating a cholinergic deficit in Alzheimer's disease due to increased butyrylcholinesterase activity [34]. Therefore, compound 5 and 6 may serve as a potential lead compound for development of a new class of drug for prevention of the progression of neurodegenerative [35].

### Conclusion

It was discovered that the synthetic approach employed for the synthesis of the various 3-methylquinolin-2-yl-hydrazones in moderate to excellent yield is highly efficient and successful. All the compounds were found to be highly selective to acetylcholinesterase and butyrylcholinesterase, and by contrast increased activity of butyrylcholinesterase, respectively. Even though their potencies are much lower than eserine, they can be used as starting lead compounds for further optimization by using the docking study on the crystal structures of the cholinesterase enzymes.

### Acknowledgements

The Authors would like to thank Professor Reuben H. Simoyi Research Group Laboratory, Portland state University, Oregon, United States of America for supporting this research.

### Table 1: IC_{50} Values for Inhibiting Activities of compounds 2-12 on Cholinesterase Enzymes. A selectivity for AChE is defined as IC_{50}(BChE)/IC_{50}(AChE), b Selectivity for BChE is defined as IC_{50}(AChE)/IC_{50}(BChE).

<table>
<thead>
<tr>
<th>Sample</th>
<th>AChE inhibition, IC_{50} (µg/mL)</th>
<th>BChE inhibition, IC_{50} (µg/mL)</th>
<th>Selectivity for AChE</th>
<th>Selectivity for BChE</th>
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References


